solution of known concentration containing approximately 250 micrograms of fluorometholone acetate per milliliter. Mix well.

(B) *Sample solution*. Shake vial thoroughly, to homogenize its contents, and immediately remove an accurately measured representative portion from it. Quantitatively dilute the suspension thus obtained with sufficient acetonitrile to obtain a solution containing 250 micrograms of fluorometholone acetate per milliliter (estimated). For instance, dilute a 1.0 milliliter aliquot of suspension with 3.0 milliliters of acetonitrile and filter.

(C) *Resolution test solution*. Prepare as directed in paragraph (b)(2)(ii)(A) of this section, except use 10 milligrams of fluorometholone in addition to the 25 milligrams of fluorometholone acetate working standard.

(iii) *System suitability requirements—* (A) *Asymmetry*. The asymmetry (A_S) is satisfactory if it is not more than 1.35 at 10 percent of peak height.

(B) *Efficiency of the column*. The efficiency of the column (h_r) is satisfactory if it is not greater than 20, equivalent to 1,000 plates for a 10-centimeter column of 5 microns or 2,500 plates for a 25-centimeter column of 5 micron size particles.

(C) *Resolution*. The resolution (R_S) between the peaks of fluorometholone acetate and fluorometholone is satisfactory if it is not less than 2.0.

(D) *Capacity factor*. The capacity factor (k) for fluorometholone acetate is satisfactory if it is in the range between 1.0 and 5.0.

(E) *Coefficient of variation*. The coefficient of variation (*RSD* in percent) of 5 replicate injections is satisfactory if it is not more than 2.0 percent. If the system suitability requirements have been met, then proceed as described in §436.216(b) of this chapter.

(iv) *Calculations*. Calculate the fluorometholone acetate content of the container as follows:

$$\begin{array}{c}\text{Milligrams of} \\ \text{fluorometholone} \\ \text{acetate} \\ \text{per container}\end{array} = \frac{A_U \times P_s \times d}{A_s \times 1{,}000}$$

A_U=Area of the fluorometholone acetate peak in the chromatogram of the sample (at a retention time equal to that observed for the standard);

A_S=Area of the fluorometholone acetate peak in the chromatogram of the fluorometholone acetate working standard;

P_S=Fluorometholone acetate content in the fluorometholone acetate working standard solution in micrograms per milliliter; and

d = Dilution factor of the sample.

(3) *Sterility*. Proceed as directed in §436.20 of this chapter, using the method described in §436.20(e)(1).

(4) *pH*. Proceed as directed in §436.202 of this chapter, using the undiluted suspension.

(5) *Tobramycin identity*. Proceed as directed in §436.318 of this chapter, except prepare the sample for assay as follows: Decant 1.0 milliliter of the unshaken sample into a test tube. Add 100 milligrams of sodium sulfate to the test tube and shake until the sodium sulfate has been dispersed. Centrifuge to obtain a clear supernatant. Use the supernatant as the sample solution.

(6) *Fluorometholone acetate identity*. The high performance liquid chromatogram of the sample determined as directed in paragraph (b)(2) of this section, compares qualitatively to that of the fluorometholone acetate working standard.

[58 FR 26671, May 4, 1993]

Subpart E—Otic Dosage Forms

§444.442 Neomycin sulfate otic dosage forms.

§§444.442a—444.442c [Reserved]

§444.442d Neomycin sulfate ointment; neomycin sulfate- ————— ointment (the blank being filled in with the established name(s) of certain other active ingredient(s)).

The requirements for certification and the tests and methods of assay for neomycin sulfate ointment and for neomycin sulfate- ————— ointment are described in §444.542a.